IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Prior Application:	T. MURAMATSU et al Serial No. 09/313,637 Filed: May 18, 1999
Group Art Unit:	1656
Examiner:	J. Taylor
For:	PREPARATION METHOD OF NUCLEIC ACID SAMPLE FOR RARE EXPRESSED GENES AND ANALYZING METHOD USING THE PREPARED NUCLEIC ACID SAMPLES THEREBY

PRELIMINARY AMENDMENT

Assistant Commissioner of Patents
Washington, D.C. 20231

Sir:

Prior to examination, please amend the above-identified application as follows:

IN THE CLAIMS

Cancel claims 1-21, and add new claims 22-27 as follows:

--22. A method for preparation of RNA sample including rare expressed genes, comprising the steps of:

(1) hybridizing an RNA having one or plural abundant expressed genes with probes, said abundant expressed genes each having a known sequence, said probes hybridizing specifically with the known sequences of said abundant expressed genes;

(2) recovering rare expressed genes not hybridized with the probes.

--23. A method for preparation of RNA sample including rare expressed genes, comprising the steps of:

(1) hybridizing an RNA having one or plural abundant expressed genes with probes, said abundant expressed genes each having a known sequence, said probes hybridizing specifically with the known sequences of said abundant expressed genes;

(2) removing said abundant expressed genes hybridized with the probes; and

(3) recovering rare expressed genes not hybridized with the probes.

--24. A method for preparation of RNA sample including rare expressed genes, comprising the steps of:

(1) hybridizing an RNA having one or plural abundant expressed genes with probes, said abundant expressed genes;

(2) digesting one or plural sequence regions of said abundant expressed genes by Ribonuclease H, the probes being specifically hybridized to the sequence regions;

(3) inactivating Ribonuclease H in a reaction solution in the step (2); and

(4) removing the probes with DNase from a reaction solution in the step (3);

wherein said abundant expressed genes hybridized with the probes are removed and rare expressed genes not hybridized with the probes are recovered.

--25. A method for preparation of cDNA sample including rare expressed genes, comprising the steps of:

(1) hybridizing mRNAs each having one or plural abundant expressed genes with probes, said abundant expressed genes each having a known sequence, said probes hybridizing specifically with the known sequences of said abundant expressed genes;

(2) digesting one or plural sequence regions of said abundant expressed genes by Ribonuclease H, the probes being specifically hybridized to the sequence regions;

(3) inactivating Ribonuclease H in a reaction solution in the step (2);

(4) removing the probes with DNase from a reaction solution in the step (3);

(5) performing cDNA synthesis reaction using said abundant expressed genes and rare expressed genes not hybridized with the probes as template, and using an oligo dT as primer, in a solution in which the probes are removed; and

(6) removing mRNAs by treating a solution in the step (5) with RNase;

wherein cDNAs originating from said rare expressed genes and not containing cDNAs originating from said abundant expressed genes are generated.

--26. A method for preparation of cDNA sample preparation including rare expressed genes, comprising the steps of:

(1) hybridizing mRNAs each having one or plural abundant expressed genes with probes, said abundant expressed

genes each having a known sequence, said probes hybridizing specifically with the known sequences in the vicinity of 3' end of said abundant expressed genes, and 3' end of said probes being modified to inhibit syntheses of cDNAs from said abundant expressed genes;

(2) performing cDNA synthesis reaction by adding an oligo dT as primer in reaction solution in the step (1), and using said abundant expressed genes and rare expressed genes not hybridized with the probes as template; and

(3) removing mRNAs by treating a solution in the step (2) with RNase;

wherein cDNAs originating from said rare expressed genes and not containing cDNAs originating from said abundant expressed genes are generated.

--27. A method for preparation of cDNA sample including rare expressed genes, comprising the steps of:

(1) adding concurrently an oligo dT as primer and probes hybridizing with mRNAs each having one or plural abundant expressed genes, into a solution containing mRNA, and allowing a reaction condition to meet hybridization condition under which the probes hybridize with mRNAs, and then performing cDNA synthesis reaction using the primer and using said abundant expressed genes and rare expressed genes not hybridized with the probes as template, wherein said abundant expressed genes have a known sequence, said probes hybridize specifically with the known sequences in the vicinity of 3'

end of said abundant expressed genes, and 3' end of said probes is modified to inhibit syntheses of cDNAs from said abundant expressed genes; and

(2) removing mRNAs by treating a solution in the step (1) with RNase;

wherein cDNAs originating from said rare expressed genes and not containing cDNAs originating from said abundant expressed genes are generated.--

REMARKS

Examination is respectfully requested.

Respectfully submitted,

John R. Mattingly
Registration No. 30,293
Attorney for Applicants

MATTINGLY, STANGER & MALUR, P.C.
104 East Hume Avenue
Alexandria, Virginia 22301
(703) 684-1120
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